

CLAIMS

We claim:

1. An aqueous mixture for inactivation of bacteria comprising:
 - (i) an additive solution wherein chloride ion, if present, is at a concentration of less than about 10 mM,
 - (ii) a biological material suspected of containing the bacteria; and
 - (iii) a pathogen inactivation compound in an amount sufficient to inactivate at least 1 log of the bacteria.
2. The aqueous mixture of claim 1 wherein the additive solution is essentially free of chloride ions.
3. The aqueous mixture of claim 1 wherein the additive solution is hypotonic.
4. The aqueous mixture of claim 1 wherein the bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, *Salmonella Typhimurium*, *Salmonella choleraesuis*, *Escherichia coli* K12, *Pseudomonas aeruginosa*, *Serratia liquifaciens*, and *Staphylococcus epidermidis*.
5. The aqueous mixture of claim 1 wherein the bacteria is a Gram negative bacteria.
6. The aqueous mixture of claim 5 wherein the Gram negative bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, *Salmonella Typhimurium*, *Salmonella choleraesuis*, *Escherichia coli* K12, *Pseudomonas aeruginosa*, and *Serratia liquifaciens*.
7. The aqueous mixture of claim 6 wherein the Gram negative bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, and *Salmonella Typhimurium*.
8. The aqueous mixture of claim 1 wherein the biological material comprises a blood product.
9. The aqueous mixture of claim 8 wherein the blood product further comprises red blood cells.

10. The aqueous mixture of claim 1 wherein the pathogen inactivation compound is more reactive at physiological pH than at a pH of about 4.
11. The aqueous mixture of claim 1 wherein the pathogen inactivation compound in the aqueous mixture is at a concentration of between about 0.1 μ M to about 5 mM.
12. The aqueous mixture of claim 11 wherein the pathogen inactivation compound is at a concentration of between about 10 μ M to about 750 μ M.
13. A method of inactivating a bacteria in a biological material suspected of containing the bacteria comprising:
 - (i) contacting the biological material with an additive solution comprising a chloride concentration of less than about 10 mM,
 - (ii) contacting the biological material with a pathogen inactivation compound in an amount sufficient to inactivate at least 1 log of the bacteria, and
 - (iii) incubating the biological material contacted with the additive solution and the pathogen inactivation compound for sufficient time to inactivate at least 1 log of the bacteria.
14. The method of claim 13 wherein the additive solution is essentially free of chloride ions.
15. The method of claim 13 wherein the additive solution is hypotonic.
16. The method of claim 13 wherein the bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, *Salmonella Typhimurium* *Salmonella choleraesuis*, *Escherichia coli* K12, *Pseudomonas aeruginosa*, and *Serratia liquifaciens*.
17. The method of claim 13 wherein the bacteria is a Gram negative bacteria.
18. The method of claim 17 wherein the Gram negative bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, *Salmonella Typhimurium* *Salmonella choleraesuis*, *Escherichia coli* K12, *Pseudomonas aeruginosa*, and *Serratia liquifaciens*.

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(iii) incubating the biological material contacted with the first additive solution and the pathogen inactivation compound for sufficient time to inactivate at least 1 log of the bacteria.

28. The method of claim 27 wherein the additive solution is hypotonic.

29. The method of claim 27 wherein the bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, *Salmonella Typhimurium* *Salmonella choleraesuis*, *Escherichia coli* K12, *Pseudomonas aeruginosa*, and *Serratia liquifaciens*.

30. The method of claim 27 wherein the bacteria is a Gram negative bacteria.

31. The method of claim 30 wherein the Gram negative bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, *Salmonella Typhimurium* *Salmonella choleraesuis*, *Escherichia coli* K12, *Pseudomonas aeruginosa*, and *Serratia liquifaciens*.

32. The method of claim 31 wherein the Gram negative bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, and *Salmonella Typhimurium*.

33. The method of claim 27 wherein the biological material comprises a blood product.

34. The method of claim 33 wherein the blood product further comprises red blood cells.

35. The method of claim 27 wherein the pathogen inactivation compound is more reactive at physiological pH than at a pH of about 4.

36. The method of claim 27 wherein the pathogen inactivation compound is at a concentration of between about 0.1 μM to about 5 mM at the beginning of said incubation.

37. The method of claim 36 wherein the pathogen inactivation compound is at a concentration of between about 10 μM to about 750 μM at the beginning of said incubation.

38. The method of claim 27 wherein the incubation is carried out at a temperature of between about 18 °C to 25 °C.
39. The method of claim 38 wherein the incubation is carried out for between about 1 hour to about 48 hours.
40. A method of inactivating a bacteria in a biological material suspected of containing the bacteria comprising:
- (i) contacting the biological material with a pathogen inactivation compound in an amount sufficient to inactivate at least 1 log of the bacteria and an additive solution comprising a chloride concentration of less than about 10 mM, and
 - (ii) incubating the biological material contacted with the additive solution and the pathogen inactivation compound for sufficient time to inactivate at least 1 log of the bacteria.
41. The method of claim 40 wherein the additive solution is essentially free of chloride ions.
42. The method of claim 40 wherein the additive solution is hypotonic.
43. The method of claim 40 wherein the bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, *Salmonella Typhimurium* *Salmonella choleraesuis*, *Escherichia coli* K12, *Pseudomonas aeruginosa*, and *Serratia liquifaciens*.
44. The method of claim 40 wherein the bacteria is a Gram negative bacteria.
45. The method of claim 44 wherein the Gram negative bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, *Salmonella Typhimurium* *Salmonella choleraesuis*, *Escherichia coli* K12, *Pseudomonas aeruginosa*, and *Serratia liquifaciens*.
46. The method of claim 45 wherein the Gram negative bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, and *Salmonella Typhimurium*.
47. The method of claim 40 wherein the biological material comprises a blood product.

48. The method of claim 47 wherein the blood product further comprises red blood cells.
49. The method of claim 40 wherein the pathogen inactivation compound is more reactive at physiological pH than at a pH of about 4.
50. The method of claim 40 wherein the pathogen inactivation compound is at a concentration of between about 0.1 μ M to about 5 mM at the beginning of said incubation.
51. The method of claim 50 wherein the pathogen inactivation compound is at a concentration of between about 10 μ M to about 750 μ M at the beginning of said incubation.
52. The method of claim 40 wherein the incubation is carried out at a temperature of between about 18 °C to 25 °C.
53. The method of claim 52 wherein the incubation is carried out for between about 1 hour to about 48 hours.
54. A method of inactivating a bacteria in a biological material suspected of containing the Gram negative bacteria comprising:
 - (i) contacting the biological material with a first additive solution which is essentially chloride free and a pathogen inactivation compound in an amount sufficient to inactivate at least 1 log of the bacteria, wherein the pathogen inactivation compound has a greater inactivation efficiency against *Yersinia enterocolitica* when used with said first additive solution than when used with a second additive solution, said second additive solution comprising at least about 10 mM chloride ion; and
 - (iii) incubating the biological material contacted with the first additive solution and the pathogen inactivation compound for sufficient time to inactivate at least 1 log of the bacteria.
55. The method of claim 54 wherein the additive solution is hypotonic.
56. The method of claim 54 wherein the bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*,

Salmonella Typhimurium Salmonella choleraesuis, Escherichia coli K12, Pseudomonas aeruginosa, and Serratia liquifaciens.

57. The method of claim 54 wherein the bacteria is a Gram negative bacteria.

58. The method of claim 57 wherein the Gram negative bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, *Salmonella Typhimurium Salmonella choleraesuis*, *Escherichia coli K12*, *Pseudomonas aeruginosa*, and *Serratia liquifaciens*.

59. The method of claim 58 wherein the Gram negative bacteria is selected from the group consisting of *Yersinia enterocolitica*, *Pseudomonas fluorescens*, *Serratia marcescens*, and *Salmonella Typhimurium*.

60. The method of claim 54 wherein the biological material comprises a blood product.

61. The method of claim 60 wherein the blood product further comprises red blood cells.

62. The method of claim 54 wherein the pathogen inactivation compound is more reactive at physiological pH than at a pH of about 4.

63. The method of claim 54 wherein the pathogen inactivation compound is at a concentration of between about 0.1 μM to about 5 mM at the beginning of said incubation.

64. The method of claim 63 where in the pathogen inactivation compound is at a concentration of between about 10 μM to about 750 μM at the beginning of said incubation.

65. The method of claim 54 wherein the incubation is carried out at a temperature of between about 18 $^{\circ}\text{C}$ to 25 $^{\circ}\text{C}$.

66. The method of claim 65 wherein the incubation is carried out for between about 1 hour to about 48 hours.

67. A method of inactivating a bacteria in a biological material suspected of containing the bacteria comprising:

- (i) contacting the biological material with an additive solution that is essentially free of chloride ions and comprises about 26.6 mM sodium citrate, about 17 mM disodium phosphate, about 4.7 mM monosodium phosphate, about 1.6 mM adenine and about 42.5 mM mannitol,
- (ii) contacting the biological material with a pathogen inactivation compound in an amount sufficient to inactivate at least 1 log of the bacteria, and
- (iii) incubating the biological material contacted with the additive solution and the pathogen inactivation compound for sufficient time to inactivate at least 1 log of the bacteria.